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Article

Fatal Diffuse Alveolar Hemorrhage (DAH) Secondary to Monoclonal Antibody Therapy in a Young Patient with Ulcerative Colitis (UC): An Autopsy Case

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Abstract: Pulmonary complications are frequently observed in patients with ulcerative colitis (UC), with large airway disease being the most prevalent manifestation. In this case study, we explore the potential for Immunosuppressant, a monoclonal antibody, to induce pulmonary hemorrhage—a side effect not previously documented with its use. The subject, a young male in his twenties with a six-year history of UC, tragically died suddenly. Found in a pool of blood without external injuries, extensive post-mortem examinations were required to determine the cause of death. The patient had been on biologic therapy, receiving Immunosuppressant intermittently over the past two years, including an infusion five days prior to death. External examination revealed bloody, pinkish foamy material expelling from his nose and mouth. An autopsy supported an atypical presentation of UC, while histopathology confirmed diffuse pulmonary hemorrhage, marked by edema and emphysematous changes. Additionally, there was a notable lymphomonocytic infiltration around the arterial walls, and signs of bronchiolitis and focal pleural fibrosis were evident. We concluded that the cause of death was Diffuse Alveolar Hemorrhage (DAH), directly associated with the recent administration of Immunosuppressant. This finding underscores the need for vigilant pulmonary monitoring in patients treated with monoclonal antibodies to promptly identify and manage potential fatal adverse reactions.

Keywords: forensic pathology; inflammatory bowel diseases; immunosuppressant; diffuse alveolar hemorrhage; monoclonal antibody; adverse reaction

1. Introduction

Ulcerative Colitis (UC) is a chronic inflammatory disorder of the large intestine that is immune-mediated. Characterized primarily by inflammation starting in the rectum and potentially extending proximally to involve sections or the entirety of the colon, UC can manifest at any age. However, it predominantly occurs in individuals, with a peak onset age between 15 and 30 years. This age-related distribution underscores the significant impact UC can have on younger populations, necessitating a focused approach to management and treatment [1]. Chronic Inflammatory Bowel Disease (IBD) encompasses two principal subtypes of persistent inflammatory conditions of the gastrointestinal tract: Crohn's Disease (CD) and Ulcerative Colitis (UC). Additionally, there exists a category termed IBDU—Inflammatory Bowel Disease Unclassified. Studies estimate that approximately 80% of IBDU cases are reclassified as either CD or UC within an eight-year period. Nevertheless, a distinct subset remains permanently diagnosed as IBDU, constituting a unique clinical entity. Long-term follow-up studies indicate that reclassification rates vary from 5-14%, with a higher incidence of transitioning to Crohn's Disease among adults [2]. The diagnosis and sub-classification of Inflammatory Bowel Disease (IBD) in clinical practice present significant challenges, often leading to diagnostic

inaccuracies. Although ileo-colonoscopy remains the gold standard for diagnosis, a multi-disciplinary approach that integrates clinical evaluation with endoscopic, histological, radiological, and biochemical investigations is essential. IBD affects not only the gastrointestinal tract but also has systemic implications, causing extra-intestinal manifestations that can impact virtually any organ system. These manifestations are a primary source of morbidity and disability. Commonly affected areas include the articular, hepato-biliary, and cutaneous districts. The probability of developing extra-intestinal manifestations escalates with the duration of the intestinal disease and is more prevalent in Crohn's disease than in Ulcerative Colitis (71% vs 21-22%). While some extra-intestinal manifestations correlate directly with the extent of intestinal inflammation, others, such as those affecting the cardiothoracic district, occur independently of the inflammatory activity within the intestines [3–6]. Respiratory complications in Inflammatory Bowel Disease (IBD) are relatively uncommon. The most frequent manifestations involve the larger airways, such as chronic bronchitis and bronchiectasis, although all lung parenchyma can be affected. These pulmonary manifestations typically occur in patients with long-standing IBD and are not directly related to exacerbations of the intestinal condition. Importantly, lung involvement can occur even in the absence of overt clinical symptoms.

Pulmonary manifestations can be categorized based on their underlying mechanisms into several types: drug-induced lung disease (associated with medications such as sulfasalazine, mesalazine, methotrexate, and infliximab), anatomical disease (such as fistulas), overlap syndromes, and immune-mediated processes. The range of pulmonary symptoms in IBD is broad, with patients potentially experiencing progressive respiratory symptoms like breathlessness, chest pain, and cough, or exhibiting changes on radiographic imaging. Interestingly, drugs like sulfasalazine and mesalazine may induce asymptomatic lung lesions more frequently than previously recognized. Typically, symptoms manifest within 2-6 months of initiating therapy; however, in some instances, they can appear within just a few days or, alternatively, after many years [7,8].

The medical management of Inflammatory Bowel Disease (IBD) involves pharmacological interventions that either directly or indirectly modulate the inflammatory cascade. Effective drugs for Ulcerative Colitis include mesalazine, corticosteroids, and immunosuppressants such as cyclosporine and azathioprine. Additionally, monoclonal antibodies targeting TNF-alpha (e.g., infliximab) are integral to the therapeutic arsenal. Immunosuppressants represent a relatively recent therapeutic option for treating patients with moderate-to-severe forms of Ulcerative Colitis and Crohn's Disease. These drugs are particularly valuable for individuals who are refractory or intolerant to conventional treatments, including anti-TNF α agents. This approach underscores the evolving landscape of IBD treatment, which increasingly prioritizes tailored therapies based on individual patient responses and tolerability profiles.

Immunosuppressants, specifically the newer class of integrin antagonist biological agents, have been introduced for treating patients with moderate-to-severe Crohn's Disease (CD) and Ulcerative Colitis (UC). These agents are particularly useful for patients who have not responded adequately to traditional therapies, including tumor necrosis factor antagonists (TNF- α). These treatments display varying advantages and response rates, highlighting the necessity for personalized medical strategies based on individual patient profiles. On the other hand, surgical therapy presents a distinct approach, often considered when medical management does not achieve sufficient control of symptoms or in cases of complications [3,9].

2. Case Presentation

A young man in his third decade of life with a history of ulcerative colitis tragically died suddenly following physical exertion. Despite emergency resuscitation attempts, he was pronounced deceased at the scene. At the time of discovery, his body was surrounded by a substantial pool of blood and foamy, reddish fluid, which had emanated from his oral and/or nasal cavities. Notably, there were no signs of external trauma, though point abrasions from resuscitation efforts were visible on his torso. A blister pack of Mesavanacol was found in his suitcase.

The deceased was subsequently transferred to the Institute of Forensic Medicine in Palermo to further the judicial investigation. An autopsy conducted three days post-mortem revealed no signs of internal bleeding within the peritoneal cavity or gastrointestinal tract, including the absence of erosions or bleeding in the stomach and the large intestine, which also showed no signs of ulcers, perforations, or bleeding.

Significant findings included a foamy, blood-stained fluid within the tracheal lumen, glottal edema, and the presence of a similar frothy, reddish liquid upon manipulation of the bronchi. The lungs were congested and markedly hemorrhagic, exuding abundant frothy material upon sectioning. These findings suggest a pulmonary cause of death, potentially linked to an acute exacerbation or complication related to his underlying ulcerative colitis, though further investigation would be necessary to confirm the exact pathophysiological mechanism (Figure 1 and 2).

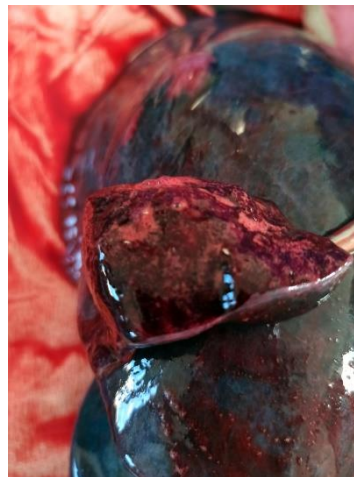


Figure 1. Particular of pulmonary edema.



Figure 2. Another focus on pulmonary edema.

The intestinal mucosa was taken every 5 cm.

Histological investigations provided significant insights into the pathological conditions affecting various organs. The large intestine exhibited chronic mucosal inflammation with focal inflammation of the submucosa, coupled with ulcerative phenomena. There was also evidence of chronic inflammation within the ileal mucosa. Notably, the heart displayed foci of active lymphocytic myocarditis in the left ventricle and diffuse colliquative myocytolysis, indicating severe cardiac tissue disruption.

Furthermore, the liver was affected by chronic lymphocytic portitis, highlighting an inflammatory response within the portal areas. At the pulmonary level, the findings were particularly

severe, with widespread pulmonary hemorrhage and areas of edema. Additionally, emphysematous changes were noted, suggesting alterations in lung tissue structure and function. These comprehensive histological findings underscore the multi-organ impact of the underlying disease processes, contributing to a complex clinical presentation. (Figures 3 and 4).

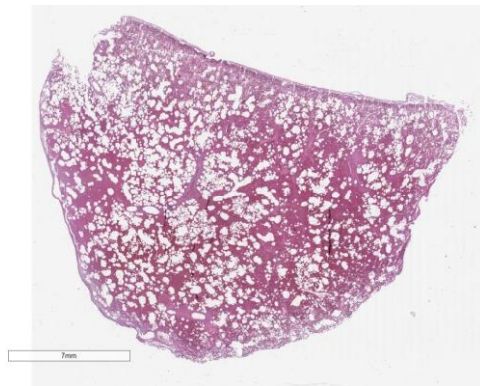


Figure 3. Section in H&E that show the hemorrhage and the alveolar damage.

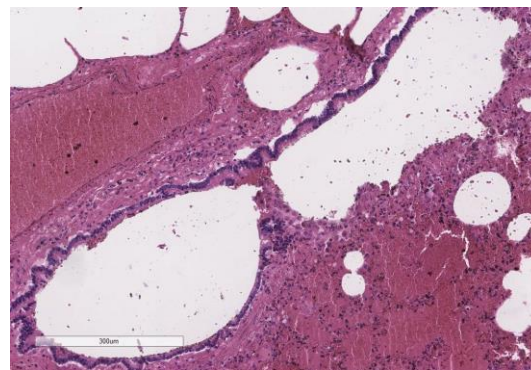


Figure 4. Section in H&E (x20) that focus on alveolar damage, and also show the hemorrhage and the infiltrate.

Furthermore, a lymphomonocytic infiltrate (Figure 5) with a periarteriolar sheath invades the vessel wall with dissociation of the tunica media, and in some tract rupture of alveolar walls. Areas of bronchiolitis, focal pleural fibrosis associated with fibroids of the central lobular and peribronchoacinar interlobar connective tissue.

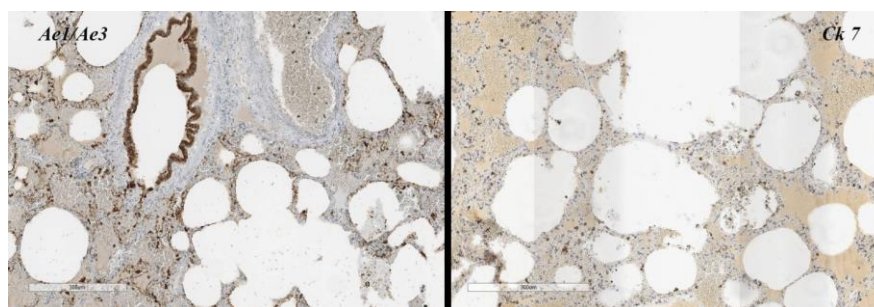


Figure 5. On the left an immunohistochemical section ck7 showing the presence of lymphocytic infiltrate, while on the right an immunohistochemical section on the Ae1/Ae3 cytokeratins showing the damage on the walls.

Toxicological analyses were conducted to screen for a spectrum of substances including opioids, benzodiazepines, cannabinoids, cocaine, and amphetamines, among others. All tests returned negative results, indicating no presence of these drugs in the system at the time of testing.

The clinical history indicated that the patient was diagnosed with ulcerative colitis five years earlier, supported by clinical symptoms, laboratory results, and endoscopic evidence. The clinical presentation included symptoms such as liquid stool discharge, abdominal pain, proctorrhagia, and weight loss. Laboratory analysis confirmed the presence of p-ANCA and PR3-cANCA antibodies, which are indicative of autoimmune activity. An ileo-colonoscopy further substantiated the diagnosis by revealing "backwash ileitis," a condition often associated with this form of inflammatory bowel disease [2].

Since his diagnosis in 2014, the patient has been under consistent clinical, laboratory, and instrumental surveillance, navigating through periods of remission and exacerbation of ulcerative colitis. Initial treatment involved topical and oral mesalazine, augmented by three cycles of steroids during acute exacerbations. In June 2019, due to insufficient response to these therapies, the patient was switched to monoclonal therapy with infliximab. However, this intervention was halted in October of the same year because of primary failure, evidenced by ongoing active disease both clinically and on ultrasound imaging. Additionally, treatment with azathioprine was terminated prior to this due to pharmacological intolerance.

In December, the patient commenced another monoclonal therapy using a different immunosuppressant, which led to clinical improvement although colonoscopy still showed persistent disease activity. The regimen for this treatment included 300 mg of the immunosuppressant administered intravenously every 8 weeks, and this frequency was increased to every 4 weeks starting in April. Oral mesalazine was continued alongside this therapy.

The patient's last chest X-ray in May 2019 was normal. Subsequent laboratory results from June 2020 indicated moderate inflammation, as evidenced by a C-reactive protein level of 118 mg/L and fecal calprotectin of 275 ng/g. A comprehensive Pancolscopy conducted on July 27, 2020, revealed colic segments with a tubular appearance. The mucosa lining these segments was hyperemic, fragile, and marked by erosions alongside minor ulcerations. Additionally, several pseudopolyps with an inflammatory appearance and eroded surfaces ranging from 0.1 to 1.5 cm were observed. The rectal ampulla also exhibited hyperemic mucosa and a loss of the vascular pattern. Tragically, on August 23, 2020, only five days following an intravenous infusion of the immunosuppressant, the young man suddenly.

3. Discussion

Immunosuppressants, while pivotal in preventing organ rejection post-transplantation and managing autoimmune diseases, can have detrimental effects on pulmonary health. These drugs can compromise the immune system's ability to combat infections, leading to an increased risk of developing severe respiratory infections such as pneumonia and tuberculosis. Furthermore, certain immunosuppressants are associated with direct toxic effects on lung tissue, which may manifest as drug-induced pneumonitis or pulmonary fibrosis. This condition involves inflammation and scarring of the lung tissue, impairing gas exchange and potentially leading to progressive and irreversible respiratory failure. The balancing act between the benefits of immunosuppression and the risk of pulmonary complications requires vigilant monitoring and management to optimize patient outcomes.

The autopsy confirmed diagnosis of RCU with multiorgan extra-intestinal manifestations (cardiac, hepatic, pulmonary) exceeded the limits of differential diagnostics. Among them was a pulmonary hemorrhage (diffuse endoalveolar).

HDA is well established in scientific literature in ANCA-associated diseases and sometimes associated with monoclonal therapy.

Excluding in the case in question all other causes of HDA (ANCA-associated diseases/vasculitis in relation to the clinical history and autopsy results), despite being present chronic lung damage from drugs, extra-intestinal respiratory manifestations from IBD or overlap exclusion [2,7], the acute cause death is due to diffuse alveolar haemorrhage secondary to Immunosuppressant as a diagnosis of exclusion [10].

Some immunosuppressant is a humanized monoclonal antibody that targets the $\alpha 4\beta 7$ -integrin expressed specifically on a subset of gut-homing T lymphocytes and inhibits its binding with mucosal addressing cell adhesion molecule 1 (MAdCAM-1). This binding of $\alpha 4\beta 7$ with MAdCAM-1 is considered gut selective. It is indicated for treatment of adult patients with active ulcerative colitis (moderate to severe) who have shown inadequate response, therapeutic failure or have been found to be intolerant to conventional therapy or administration of an antagonist of tumour necrosis factor α (TNF α)[11].

Recent literature data suggest that although immunosuppressant has a high safety profile, it takes action also on the lung. In fact, $\alpha 4\beta 7$ is also moderately expressed on other types of leukocytes and $\alpha 4\beta 7$ is known to have a small role in pulmonary homing through VCAM1 and fibronectin, with the $\alpha 4\beta 7$ - fibronectin interaction also being inhibited by immunosuppressant. However, pulmonary homing is more dependent on $\beta 1$ integrin and it has been suggested that immunosuppressant could induce a shift in integrin expression on lymphocytes with upregulation of $\beta 1$, leading to an increased migration to extraintestinal sites, thereby facilitating development of extraintestinal inflammation. There are indeed arguments for an interaction between the gastrointestinal and bronchial mucosa mediated by the immune system [8,12–15]. Common respiratory adverse reactions of Immunosuppressant reported include oropharyngeal pain, nasal congestion and coughing. The serum half-life of the drug is about 26 days; the exact route of elimination of Immunosuppressant is not known [15].

Death was caused by pulmonary hemorrhage (HDA) responsible for the massive loss of blood observed during the judicial inspection and responsible for terminal cardiac arrest (diffuse colliquative myocytosis) compatible with the infusion of Immunosuppressant 5 days before death.

The purpose of our case report is to show the possibility that Vedolizumab causes pulmonary hemorrhage such as many other monoclonal antibodies to different farmodynamics like Infliximab, Ustekinumab and others [9,18,20]. In fact, rapid onset pulmonary manifestations are known, which if caught in time can be managed safely for the patient [19,20].

The classic pulmonary manifestations of pulmonary RU are dictated by the chronic physiological mechanism of the disease.

In fact, upon self-examination, the lungs showed a rapid picture of disease, with rapid onset, and the microscopic examination highlighted a picture of diffuse pulmonary hemorrhage with areas of edema and emphysematous expressions. Furthermore, a lymphomonocytic infiltrate with a periarteriolar sheath invades the vascular wall with dissociation of the tunica media.

In relation to the action that monoclonal therapy has on the migration of the lymphocyte population, and supported by the immunohistochemistry [Figure 5] which shows a notable lymphocyte infiltrate present in the affected area, we posed the problem in relation to immunosuppressor. However, in cases like ours, in which the rapid onset does not present all the pulmonary clinical manifestations, it leads us to advise specialists to closely follow up the patient who uses this drug, also reporting minor anomalous signs (cough, fatigue, respiratory problems).

4. Conclusions

In conclusion, while immunosuppressants are indispensable for their role in controlling autoimmune disorders and preventing transplant rejection, their use is not without significant pulmonary risks. One of the most severe complications is Diffuse Alveolar Hemorrhage (DAH), a potentially life-threatening condition characterized by bleeding into the alveoli of the lungs. DAH can be precipitated by the immunosuppressive therapy weakening the capillary integrity or through immune-mediated vascular injury. Effective management of patients on immunosuppressant therapy requires a careful consideration of these risks, regular monitoring for early signs of pulmonary distress, and prompt intervention to mitigate adverse outcomes. Balancing these aspects is crucial to harnessing the benefits of immunosuppressants while minimizing their potential harm to lung health.

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